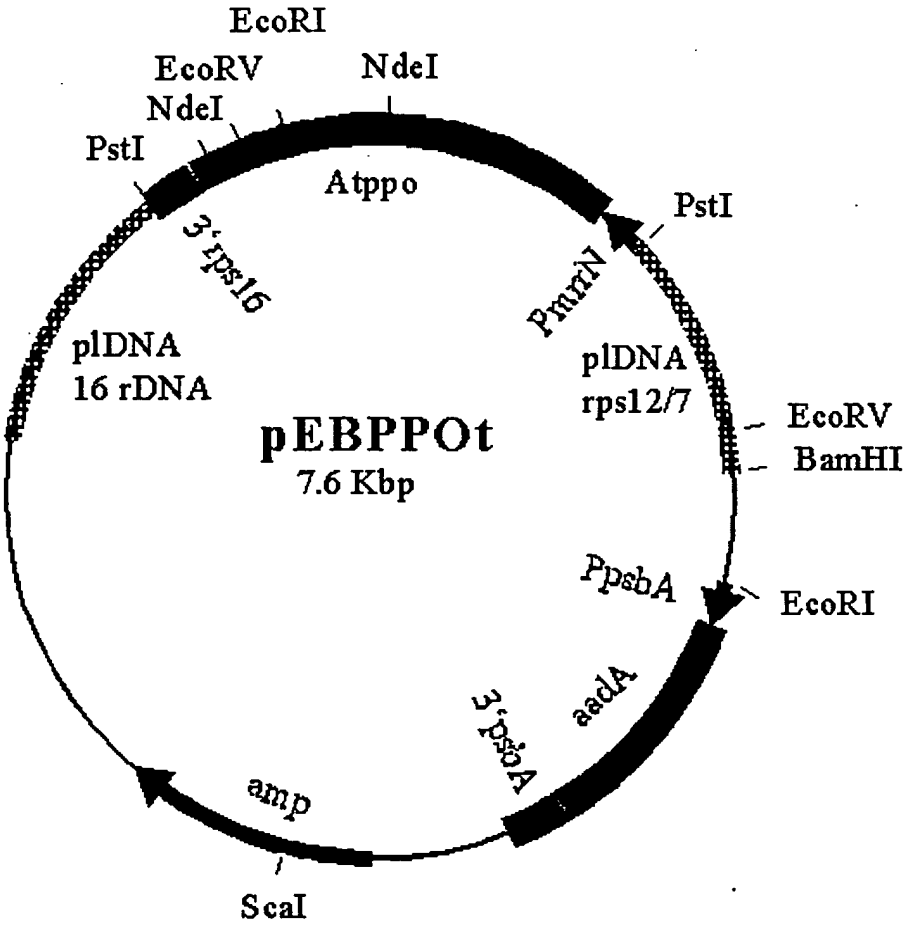
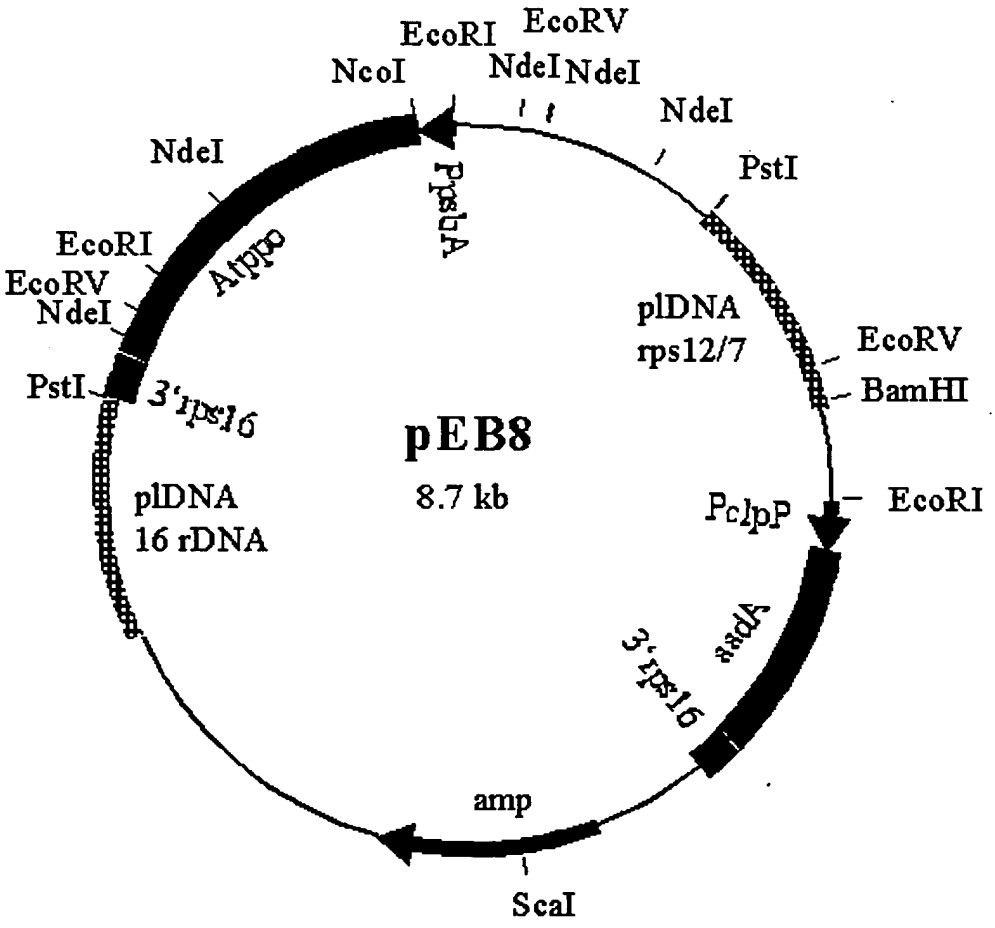
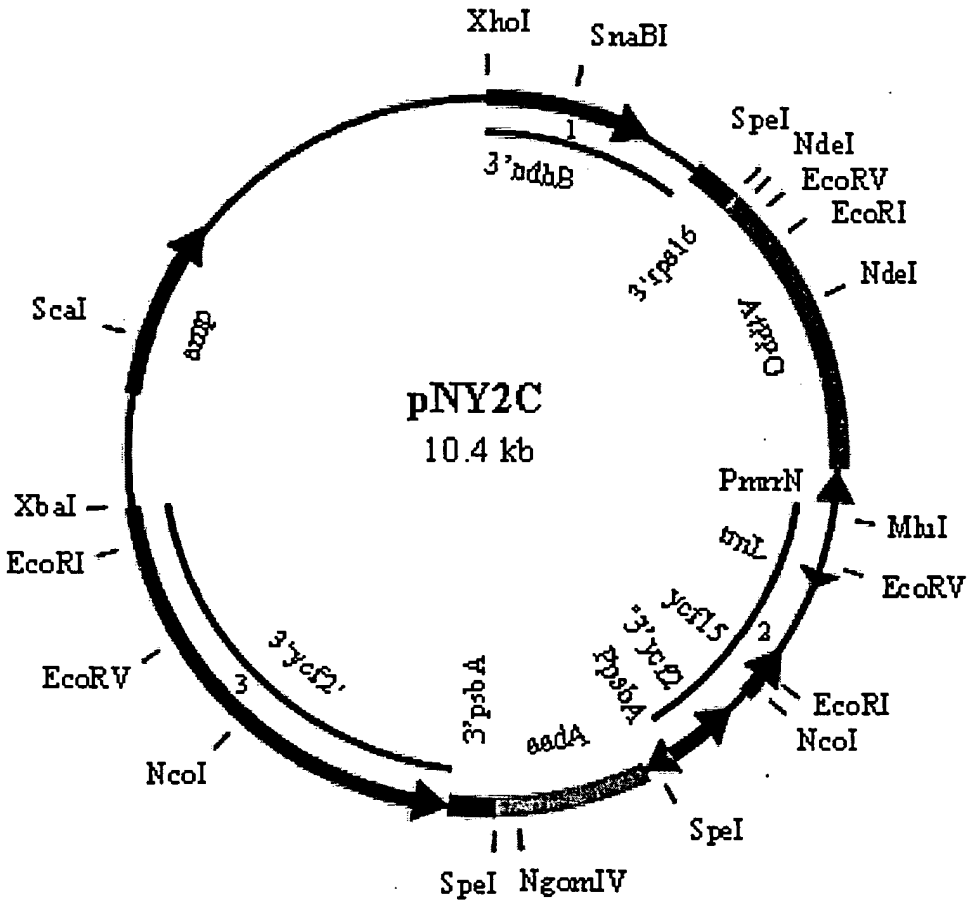
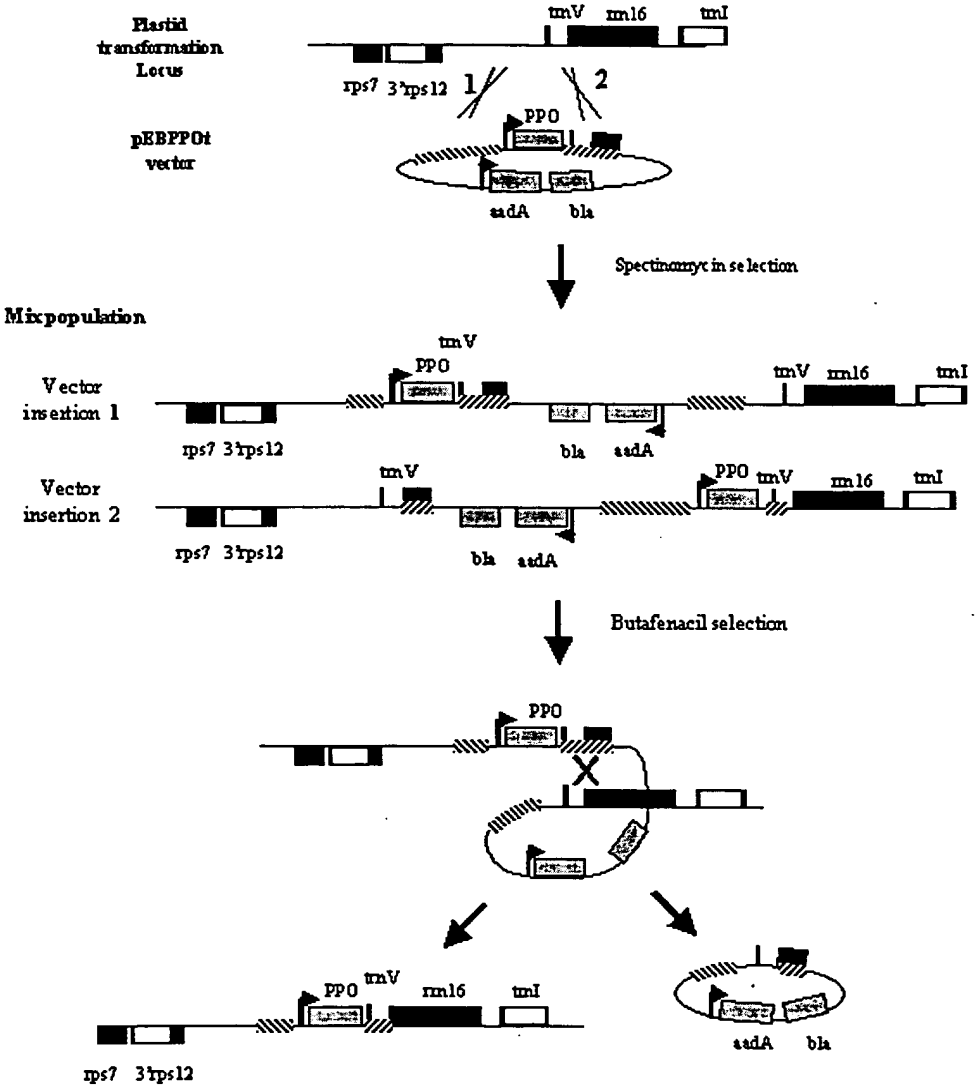
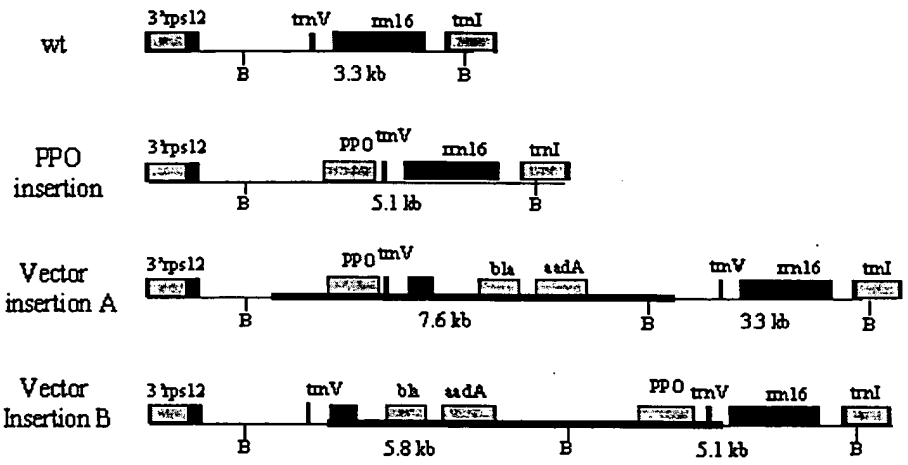

EcoRI
EcoRV
NdeI
NdeI
PstI
Atppo
PstI
3'rps16
PmrrN
plDNA
16 rDNA
plDNA
rps12/7
pEBPPOt
7.6 Kbp
EcoRV
BamHI
PpsbA
EcoRI
aadA
3'psbA
amp
ScaI


EcoRI
EcoRV
NcoI
NdeI
NdeI
NdeI
PstI
NdeI
EcoRI
EcoRV
NdeI
PstI
3'rps16
plDNA
16 rDNA
pEB8
8.7 kb
plDNA
rps12/7
EcoRV
BamHI
PclpP
EcoRI
3'rps16
amp
ScaI

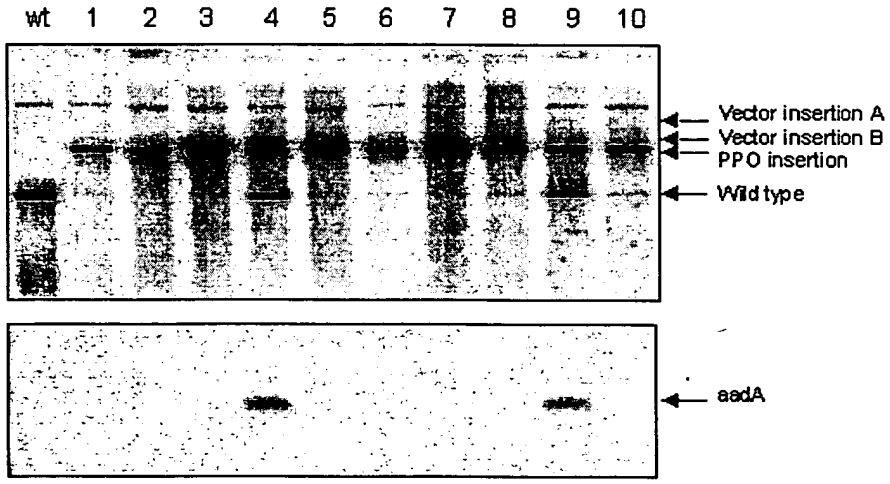

pNY2C
10.4 kb
XhoI
SnaBI
SpeI
NdeI
EcoRV
EcoRI
NdeI
MluI
EcoRV
EcoRI
NcoI
SpeI
SpeI
NgomIV
NcoI
EcoRV
EcoRI
XbaI
ScaI

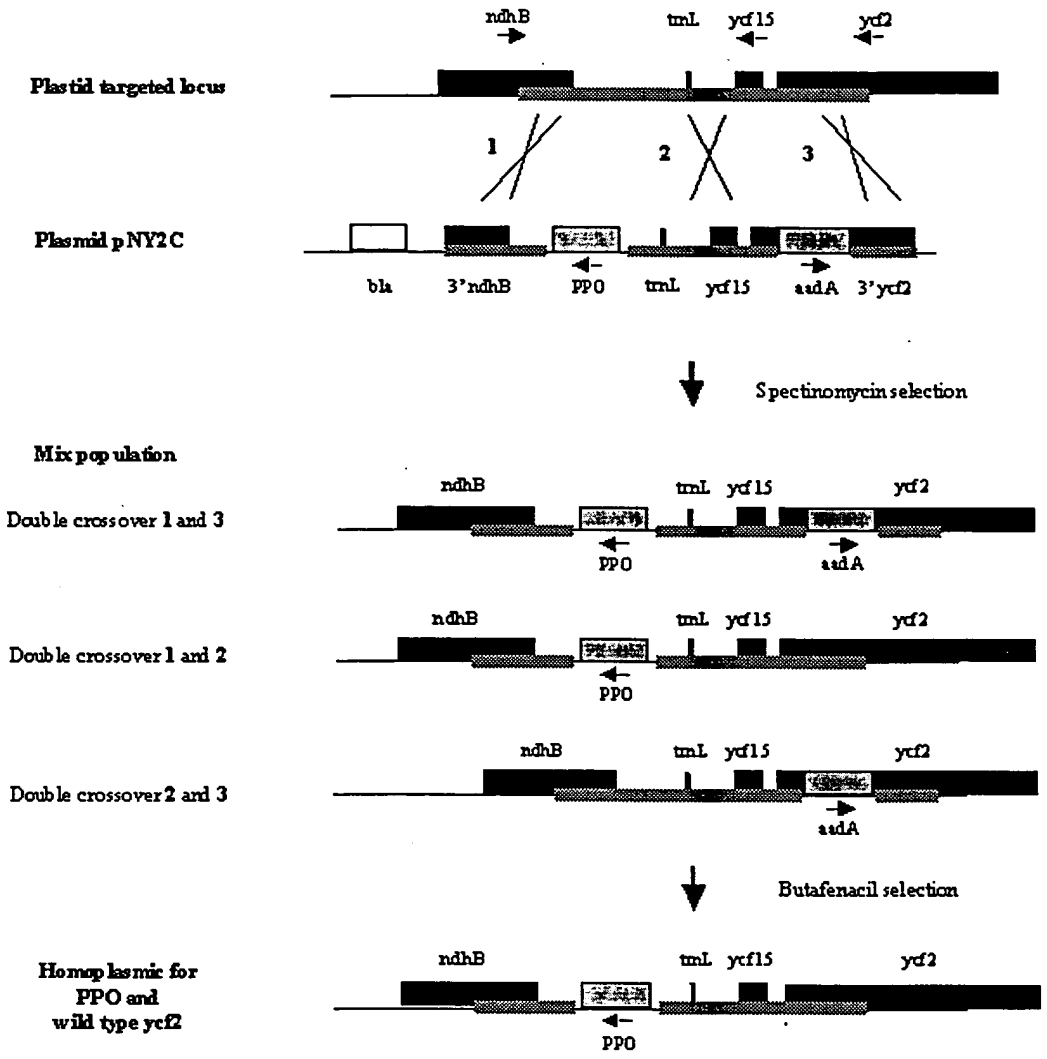

Plastid transformation Locus
pEBPPOt vector
trnV
rrn16
trnI
rps7
3'rps12
1
2
PPO
aadA
bla
Spectinomycin selection
Mixpopulation
Vector insertion 1
Vector insertion 2
Butafenacil selection

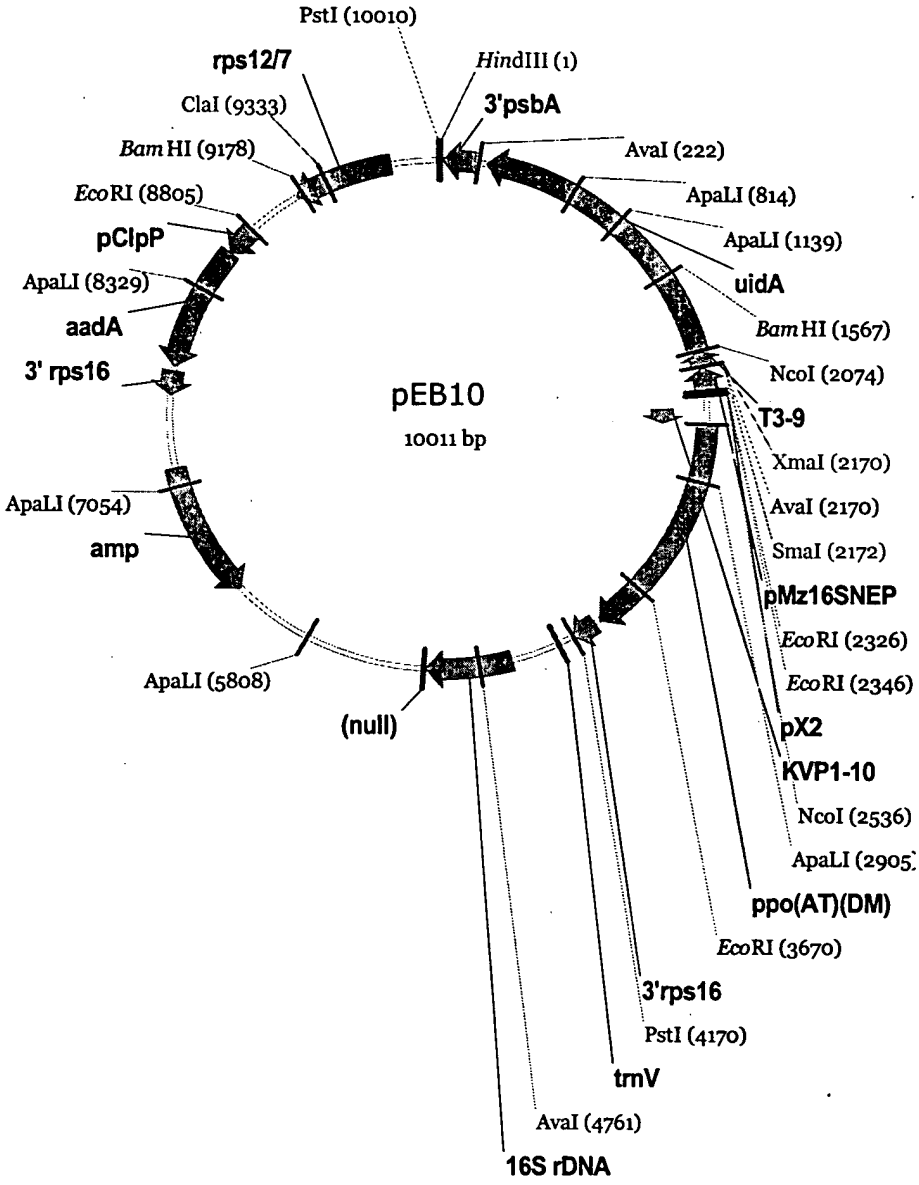
A


wt
3'rps12
trnV
rrn16
trnI
B
3.3 kb
B
PPO insertion
PPO
5.1 kb
Vector insertion A
bla
aadA
7.6 kb
3.3 kb
Vector Insertion B
5.8 kb
5.1 kb

B


wt 1 2 3 4 5 6 7 8 9 10
Vector insertion A
Vector insertion B
PPO insertion
Wild type
aadA


Plastid targeted locus
ndhB
trnL
ycf15
ycf2
1
2
3
Plasmid pNY2C
bla
3'ndhB
PPO
trnL
ycf15
aadA
3'ycf2
Spectinomycin selection
Mix population
Double crossover 1 and 3
Double crossover 1 and 2
Double crossover 2 and 3
Butafenacil selection
Homoplasmic for PPO and wild type ycf2


pEB10
10011 bp
PstI (10010)
HindIII (1)
3'psbA
AvaI (222)
ApaLI (814)
ApaLI (1139)
uidA
BamHI (1567)
NcoI (2074)
T3-9
XmaI (2170)
AvaI (2170)
SmaI (2172)
pMz16SNEP
EcoRI (2326)
EcoRI (2346)
pX2
KVP1-10
NcoI (2536)
ApaLI (2905)
ppo(AT)(DM)
EcoRI (3670)
3'rps16
PstI (4170)
trnV
AvaI (4761)
16S rDNA
(null)
ApaLI (5808)
amp
ApaLI (7054)
3' rps16
aadA
ApaLI (8329)
pClpP
EcoRI (8805)
BamHI (9178)
ClaI (9333)
rps12/7